

KITAKANTO MEDICAL JOURNAL AWARD

Stress-induced Biomarkers in Liver with Non-alcohol Fatty Liver Diseases and Non-alcohol Steatohepatitis

Hiromi Ono¹, Akira Tanaka^{1,2}, Kyoumi Nakazato³, Yutaka Hasegawa⁴, Ke Ih Kim⁵, Soo Ryang Kim⁵, Katsuyuki Nakajima^{2,3} and Takeaki Nagamine³

1 Laboratory of Clinical Nutrition and Medicine, Kagawa Nutrition University, 3-9-21 Chiyoda, Sakado, Saitama 350-0288, Japan

2 Nutrition Clinic, Kagawa Nutrition University, 3-24-3 Komagome, Toshima-ku, Tokyo 170-8481, Japan

3 Gunma University Graduate School of Health Sciences, 3-39-22 Showa-machi, Maebashi, Gunma 371-8514, Japan

4 Educational Center for Clinical Pharmacy, Kobe Pharmaceutical University, 4-19-1 Motoyamakitamachi, Higashinada-ku, Kobe, Hyogo 658-8558, Japan

5 Department of Gastroenterology, Kobe Asahi Hospital, 3-5-25 Boh-oh-ji-cho, Nagata-ku, Kobe, Hyogo 653-0801, Japan

Article Information

Publication history:

Received: December 16, 2015

Accepted: December 28 2015

Corresponding author:

Kyoumi Nakazato

Gunma University Graduate School of Health Sciences,
3-39-22 Showa-machi, Maebashi, Gunma 371-8514, Japan

Tel: +81-27-220-8977

E-mail: nkyoumi@gunma-u.ac.jp



Background

A comparative study between plasma diagnostic markers and oxidative stress-induced biomarkers localized differently in liver has not been reported in non-alcohol fatty liver (NAFLD) and non-alcoholic steatohepatitis (NASH).

Methods

Pathological observations by Hematoxylin and Eosin (HE) staining and immunostaining by specific antibodies against metallothionein (MT)-1/2 and -3, heme oxygenase -1 (HO-1), adiponectin using biopsy samples and plasma diagnostic makers were determined in 37 cases.

Results

The MT-1/2, HO-1 and adiponectin levels were all significantly reduced in the liver with NASH compared with NAFLD and control. MT-1/2 was most strongly stained in hepatocytes in the normal and NAFLD liver, while it was significantly reduced in NASH. Adiponectin was stained significantly less at

blood vessel cells in NASH compared with NAFLD and controls. HO-1 was also stained significantly less in the Kupffer cells in NASH compared with NAFLD and controls. MT-3 was stained similarly among the three groups at blood vessel cells. Those biomarkers trended negatively with plasma liver injury biomarkers.

Conclusions

The significantly reduced expression of oxidative stress-induced biomarkers in NASH may be associated with the degree of pathological damage. In particular, MT-1/2 seemed to play the important role in hepatocytes against stress-induced damage in NASH.

References

1. Matteoni CA, Younossi ZM, Gramlich T, et al. Non-alcoholic fatty liver disease: a spectrum of clinical and pathological study. *Gastroenterology* 1999; 116: 1413-1419.
2. James OFL, Day CP. Non-alcoholic steatohepatitis (NASH): a disease of emerging identity and importance. *J Hepatol* 1998; 29: 495-501.
3. Sheth SG, Gordon FD, Chopra S. Non-alcoholic steatohepatitis. *Ann Intern Med* 1997; 126: 137-145.

4. Ludwig J, McGill DB, Lindor KD. Review: non-alcoholic steatohepatitis. *J Gastroenterol Hepatol* 1997; 12: 398-403.
5. Bacon BR, Farahvash MJ, Janney CG, et al. Nonalcoholic steatohepatitis: an expanded clinical entity. *Gastroenterology* 1994; 107: 1103-1109.
6. Teri MR, James OFL, Burt AD, et al. The natural history of non-alcoholic fatty liver: a follow-up study. *Hepatology* 1995; 22: 1714-1719.
7. Powell EE, Cooksley WGE, Hanson R, et al. The natural history of non-alcoholic steatohepatitis: a follow-up study of forty-two patients for up to 21 years. *Hepatology* 1990; 11: 74-80.
8. Lee RG. Non-alcoholic steatohepatitis: a study of 49 patients. *Hum Pathol* 1989; 20: 594-598.
9. Ludwig J, Vaggiano TR, McGill DB, et al. Non-alcoholic steatohepatitis: Mayo Clinic experiences with a hitherto unnamed disease. *Mayo Clin Proc* 1980; 55: 434-438.
10. Sanyal AJ, Campbell-Sargent C, Mirshahi F, et al. Non-alcoholic steatohepatitis: association of insulin resistance and mitochondrial abnormalities. *Gastroenterology* 2001; 120: 1183-1192.
11. Weltman MD, Farrell GC, Hall P, et al. Hepatic cytochrome P450 2E1 is increased in patients with non-alcoholic steatohepatitis. *Hepatology* 1998; 27: 128-133.
12. Berson A, De Beco V, Letteron P, et al. Steatohepatitis-inducing drugs cause mitochondrial dysfunction and lipid peroxidation in rat hepatocytes. *Gastroenterology* 1998; 114: 764-774.
13. Day CP, James OFW. Steatohepatitis: a tale of two 'Hits'. *Gastroenterology* 1998; 114: 842-845.
14. Halliwell B. Free radicals, antioxidant, and human disease: curiosity, cause, or consequence? *Lancet* 1994; 344: 721-724.
15. Slater TF. Free-radical mechanisms in tissue injury. *Biochem J* 1984; 222: 1-15.
16. Mohommad MK, Zhou Z, Cave M, et al. Zinc and liver disease. *Nutr Clin Pract* 2012; 27(1): 8-20.
17. Ohtsuji M, Katsuoka F, Kobayashi A, et al. Nrf1 and Nrf2 play distinct roles in activation of antioxidant response element-dependent genes. *J Biol Chem* 2008; 283(48): 33554-33562.
18. Hozumi I, Suzuki JS, Kanazawa H, et al. Metallothionein-3 is expressed in the brain and various peripheral organs of the rat. *Neurosci Lett* 2008; 438(1): 54-58.
19. Wu BJ, Kathir K, Witting PK, et al. Antioxidants protect from atherosclerosis by a heme oxygenase-1 pathway that is independent of free radical scavenging. *J Exp Med* 2006; 203: 1117-1127.
20. Wang RQ, Nan YM, Han F, et al. The role of heme oxygenase-1 in non-alcoholic steatohepatitis. *Zhonghua Gan Zang Bing Za Zhi* 2010; 18(9): 680-684.
21. Malaguarnera L, Madeddu R, Palio E, et al. Heme oxygenase-1 levels and oxidative stress-related parameters in non-alcoholic fatty liver disease patients. *J Hepatol* 2005; 42(4): 585-591.
22. Inoue M, Tazuma S, Kanno K, et al. Bach1 gene ablation reduces steatohepatitis in mouse MCD diet model. *J Clin Biochem Nutr* 2011; 48(2): 161-166.
23. Buechler C, Wanninger J, Neumeier M. Review: Adiponectin, a key adipokine in obesity related liver diseases. *World J Gastroenterol* 2011; 17: 2801-2811.
24. Targher G, Bertolini L, Zenari L. Hypoadiponectinemia is closely associated with nonalcoholic hepatic steatosis in obese subjects. *Diabetes Care* 2004; 27: 2085-2086.
25. Targher G, Bertolini L, Rodella S, et al. Associations between plasma adiponectin concentrations and liver histology in patients with nonalcoholic fatty liver disease. *Clin Endocrinol (Oxf)* 2006; 64: 679-683.
26. Nakazato K, Nakajima K, Kusakabe T, et al. Immunohistochemical staining with newly developed metallothionein fragment antibodies against NH₂-terminal, middle-regional and COOH-terminal peptides in rabbits. *Pathol Int* 2008; 58: 765-770.
27. Nakajima K, Kodaira T, Kato M, et al. Development of an enzyme-linked immunosorbent assay for metallothionein-I and -II in plasma of humans and experimental animals. *Clin Chim Acta* 2010; 411: 758-761.
28. Nakazato K, Nakajima K, Nakano T, et al. Metallothionein (MT) 1/2 expression in MT 1/2 and MT 3 knock-out mice and Long-Evans Cinnamon (LEC) rats. *J Toxicol Sci* 2012; 37(1): 169-175.
29. Saito H, Nakazato K, Kato M, et al. Determination of metallothionein-3 by a competitive enzyme-linked immunosorbent assay in experimental animals. *J Toxicol Sci* 2013; 38: 83-91.
30. Matteoni CA, Younossi ZM, Gramlich T, et al. Non-alcoholic fatty liver disease: a spectrum of clinical and pathological severity. *Gastroenterology* 1999; 116: 1413-1419.
31. Manuel Y, Thomas Y, Pellegrini O. Review: Metallothionein and tissue damage. *IARC Sci Publ* 1992; 118: 231-237.
32. M Levrero. Viral hepatitis and liver cancer: the case of hepatitis C. *Oncogene* 2006; 25: 3834-3847.
33. Park Y, Yu E. Expression of metallothionein-1 and metallothionein-2 as a prognostic marker in hepatocellular carcinoma. *J Gastroenterol Hepatol* 2013; 28(9): 1565-1572.
34. Ratziu V, Zelber-Sagi S. Pharmacologic therapy of non-alcoholic steatohepatitis. *Clin Liver Dis* 2009; 13(4): 667-688.
35. Merat S, Aduli M, Kazemi R, et al. Liver histology changes in nonalcoholic steatohepatitis after one year of treatment with probucol. *Dig Dis Sci* 2008; 53(8): 2246-2250.
36. Tokushige K, Hashimoto E, Yatsuji S, et al. Combined pantethine and probucol therapy for Japanese patients with non-alcoholic steatohepatitis. *Hepatol Res* 2007; 37(10): 872-877.
37. Merat S, Malekzadeh R, Sohrabi MR, et al. Probuol in the treatment of non-alcoholic steatohepatitis: a double-blind randomized controlled study. *J Hepatol* 2003; 38(4): 414-418.
38. Merat S, Malekzadeh R, Sohrabi MR, et al. Probuol in the treatment of nonalcoholic steatohepatitis: an open-labeled study. *J Clin Gastroenterol* 2003; 36(3): 266-268.
39. Deng YM, Wu BJ, Witting PK, et al. Probuol protects against smooth muscle cell proliferation by upregulating heme oxygenase-1. *Circulation* 2004; 110(13): 1855-1860.
40. Li C, Hossieny P, Wu BJ, et al. Review: Pharmacologic induction of heme oxygenase-1. *Antioxid Redox Signal* 2007; 9(12): 2227-2239.
41. Delaigle AM, Senou M, Guiot Y, et al. Induction of adiponectin in skeletal muscle of type 2 diabetic mice: In vivo and in vitro studies. *Diabetologia* 2006; 49(6): 1311-1323.
42. Rong H, Tan M. Effect of probucol on serum malondialdehyde and superoxide dismutase in patients with primary hypertension. *Zhong Nan Da Xue Xue Bao Yi Xue Ban* 2012; 37(5): 458-462.
43. Li T, Danelisen I, Belló-Klein A, et al. Effects of probucol on changes of antioxidant enzymes in adriamycin-induced cardiomyopathy in rats. *Cardiovasc Res* 2000; 46(3): 523-530.